# PATENT COOPERATION TREATY

From the:
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:

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Hamilton
NEW ZEALAND

## PCT

NOTIFICATION OF TRANSMITTAL OF INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Rule 71.1)

Date of mailing
*day/month/year* 30 MAR 2004

Applicant's or agent's file reference
31378/14AJC

**IMPORTANT NOTIFICATION**

| International Application No. | International Filing Date | Priority Date |
|---|---|---|
| **PCT/NZ2003/000119** | 10 June 2003 | 10 June 2002 |

Applicant
AGRESEARCH LIMITED et al

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.

2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.

3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translations to those Offices.

4. **REMINDER**

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices)(Article 39(1))(see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide

| Name and mailing address of the IPEA/AU | Authorized officer |
|---|---|
| AUSTRALIAN PATENT OFFICE<br>PO BOX 200, WODEN ACT 2606, AUSTRALIA<br>E-mail address: pct@ipaustralia.gov.au<br>Facsimile No. (02) 6285 3929 | DAVID OLDE<br>Telephone No. (02) 6283 2569 |

# PATENT COOPERATION TREATY

# PCT

## INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)



| Applicant's or agent's file reference | FOR FURTHER ACTION | See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416). |
|---|---|---|
| 31378/14AJC | | |
| International Application No. | International Filing Date *(day/month/year)* | Priority Date *(day/month/year)* |
| PCT/NZ2003/000119 | 10 June 2003 | 10 June 2002 |

International Patent Classification (IPC) or national classification and IPC

Int. Cl.[7] C12Q 1/68 C12N 15/11 C12N 15/31

Applicant

AGRESEARCH LIMITED et al

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 5 sheets, including this cover sheet.

   [X] This report is also accompanied by ANNEXES, i.e., sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 5 sheet(s).

3. This report contains indications relating to the following items:

| | | |
|---|---|---|
| I | [X] | Basis of the report |
| II | [ ] | Priority |
| III | [ ] | Non-establishment of opinion with regard to novelty, inventive step and industrial applicability |
| IV | [ ] | Lack of unity of invention |
| V | [X] | Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement |
| VI | [X] | Certain documents cited |
| VII | [ ] | Certain defects in the international application |
| VIII | [ ] | Certain observations on the international application |

| Date of submission of the demand | Date of completion of the report |
|---|---|
| 24 November 2003 | 23 March 2004 |
| Name and mailing address of the IPEA/AU | Authorized Officer |
| AUSTRALIAN PATENT OFFICE<br>PO BOX 200, WODEN ACT 2606, AUSTRALIA<br>E-mail address: pct@ipaustralia.gov.au<br>Facsimile No. (02) 6285 3929 | **DAVID OLDE**<br>Telephone No. (02) 6283 2569 |

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No.
PCT/NZ2003/000119

## I. Basis of the report

1. With regard to the elements of the international application:*

- [ ] the international application as originally filed.
- [X] the description, pages **1-27**, as originally filed,
  pages , filed with the demand,
  pages , received on with the letter of
- [X] the claims, pages , as originally filed,
  pages , as amended (together with any statement) under Article 19,
  pages , filed with the demand,
  pages **28-32**, received on with the letter of
- [X] the drawings, pages **1/6-6/6**, as originally filed,
  pages , filed with the demand,
  pages , received on with the letter of
- [X] the sequence listing part of the description:
  pages **1-2**, as originally filed
  pages , filed with the demand
  pages , received on with the letter of

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.
These elements were available or furnished to this Authority in the following language which is:

- [ ] the language of a translation furnished for the purposes of international search (under Rule 23.1(b)).
- [ ] the language of publication of the international application (under Rule 48.3(b)).
- [ ] the language of the translation furnished for the purposes of international preliminary examination (under Rules 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- [X] contained in the international application in written form.
- [ ] filed together with the international application in computer readable form.
- [ ] furnished subsequently to this Authority in written form.
- [X] furnished subsequently to this Authority in computer readable form.
- [ ] The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- [X] The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

4. [X] The amendments have resulted in the cancellation of:

- [ ] the description, pages
- [X] the claims, Nos. **31-40**
- [ ] the drawings, sheets/fig.

5. [ ] This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).**

* *Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17).*

** *Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report*

**V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement**

1. Statement

| | | |
|---|---|---|
| Novelty (N) | Claims 1-30 | YES |
| | Claims - | NO |
| Inventive step (IS) | Claims 1-30 | YES |
| | Claims - | NO |
| Industrial applicability (IA) | Claims 1-30 | YES |
| | Claims - | NO |

2. Citations and explanations (Rule 70.7)

The invention resides in the identification of differences between the sheep and cattle *M. paratuberculosis* strains at the IS900 insertion site and the use of such differences to distinguish these strains from one another and other *M. paratuberculosis* strains. Specifically the invention resides in the sequence differences between Seq ID No 1 (sheep) and Seq ID No 2 (cattle).

Claims 1-4 relate to Seq ID No 1 and probes thereof to detect sheep *M. paratuberculosis*.

Claim 5 relates to the use of Seq ID No 2 to detect cattle *M. paratuberculosis*.

Claim 6 relates to a method of distinguishing cattle and sheep types by comparing the differences between Seq ID No 1 and Seq ID No 2.

Claims 7-20 relate to methods of detecting *M. paratuberculosis* based on the amplification of the nucleic acid of claim 1, wherein the method can comprise PCR primers based on Seq ID No 1 and/or Seq ID No 2 and/or the IS900 sequence.

Claims 21-30 relate to the use of probes based on either Seq ID No 1 or Seq ID No 2 to detect or distinguish *M. paratuberculosis* stains.

The following documents identified in the International Search report have been considered for the purposes of this report:

D1: EP1223225 (See Box VI)

D2: Coetsier *et al*, 2000

D3: Whittington *et al*, 2000

D4: Bull *et al*, 2000 and GenBank AJ250016 and GenBank AJ011838

D5: US5968815

D6: Bauerfeind *et al*, 1996

D7: EP0288306

D8: AU632383

## VI. Certain documents cited

1. Certain published documents (Rule 70.10)

| Application No. Patent No. | Publication date *(day/month/year)* | Filing date *(day/month/year)* | Priority date ( valid claim) *(day/month/year)* |
|---|---|---|---|
| EP1223225 | 17/7/02 | 10/01/01 | 10/01/01 |

EP1223225 (D1) was published after the priority date of the present application but has an earlier priority date than the current application. Thus under certain jurisdictions this citation may have become relevant at national phase as a whole of contents citation. D1 discloses a PCR detection method for *M. paratuberculosis* based on the IS900 insertion sequence. However after amendment to the claims under Article 34, this citation is no longer considered relevant to the claimed invention.

2. Non-written disclosures (Rule 70.9)

| Kind of non-written disclosure | Date of non-written disclosure *(day/month/year)* | Date of written disclosure referring to non-written disclosure *(day/month/year)* |
|---|---|---|

**Supplemental Box**
(To be used when the space in any of the preceding boxes is not sufficient)

**Continuation of Box V**

NOVELTY (N) AND INVENTIVE STEP (IS)

Claims 1-30 are considered novel and inventive in light of citations D2-D8 as none of the citations disclose or suggest SEQ ID No 1 or 2 and the use of such sequences in methods of detection or distinguishing between sheep and/or cattle *M. paratuberculosis* strains.

INDUSTRIAL APPLICABILITY(IA)

Claims 1-30 meet the requirements of the PCT in regard to industrial applicability.

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WHAT WE CLAIM IS:

1. An isolated nucleic acid molecule of a sheep type of *M. paratuberculosis* said molecule comprising SEQ ID NO. 1 or a complement thereof.

2. A probe comprising SEQ ID NO.1 or a complement thereof.

3. A probe comprising at least 20 or more contiguous nucleotides selected from nucleotides 230 – 260 of SEQ ID NO. 1 or a complement thereof.

4. The use of a nucleic acid molecule or probe as claimed in any one of claims 1-3 for detecting the presence of sheep types of *M. paratuberculosis*.

5. The use of SEQ ID NO 2 or, a fragment or complement thereof for detecting the presence of cattle types of *M. paratuberculosis*.

6. A method of distinguishing between cattle and sheep types of *M. paratuberculosis* comprising the step of comparing differences between the nucleotide sequences of SEQ ID NO. 1 and SEQ ID NO. 2 or complements of said sequences.

7. A method of detecting the presence of *M. paratuberculosis* in a sample via a nucleic acid amplification technique said method comprising the steps of:

   a) taking a sample from an animal or any other source;

   b) extracting nucleic acids from the sample or culturing mycobacteria from the sample and extracting nucleic acids from the mycobacterial culture;

c) performing a nucleic acid amplification technique with one or more nucleic acid sequences as claimed in claim 1; and

d) determining the identity of the amplification product.

8. A method as claimed in claim 7 wherein the animals may include cattle, sheep, deer, goats, ferrets, rabbits and humans.

9. A method as claimed in claim 7 wherein step d) of the method comprises identifying the presence of 10-12 contiguous nucleotides of the nucleic acid molecule comprising SEQ ID NO. 1 or a complement thereof.

10. A method claimed in claim 7 wherein step d) of the method comprises identifying the presence of at least 15 contiguous nucleotides of the nucleic acid molecule comprising SEQ ID NO. 1 or a complement thereof.

11. A method as claimed in claim 7 wherein step d) of the method comprises identifying the presence of substantially 20 contiguous nucleotides of the nucleic acid molecule comprising SEQ ID NO. 1 or a complement thereof.

12. A method as claimed in claim 7 step c) utilizes one oligonucleotide primer complementary to 10-12 contiguous nucleotides of SEQ ID NO. 1 or a complement thereof; and one oligonucleotide primer complementary to 10-12 nucleotides of IS*900* or a complement thereof.

13. A method as claimed in claim 7 wherein step c) utilizes one oligonucleotide primer complementary to substantially 15 contiguous nucleotides of SEQ ID NO. 1 or a complement thereof; and one oligonucleotide primer complementary to substantially 15 nucleotides of IS*900* or a complement thereof.

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14. A method as claimed in claim 7 wherein step c) utilizes one oligonucleotide primer complementary to substantially 20 contiguous nucleotides of SEQ ID NO. 1 or a complement thereof; and one oligonucleotide primer complementary to substantially 20 nucleotides of IS*900* or a complement thereof.

15. A method as claimed in claim 7 wherein step d) of the method comprises identifying the presence of 10-12 contiguous nucleotides of the nucleic acid molecule comprising SEQ ID NO. 2 or a complement thereof.

16. A method as claimed in claim 7 wherein step d) of the method comprises identifying the presence of at least 15 contiguous nucleotides of the nucleic acid molecule comprising SEQ ID NO. 2 or a complement thereof.

17. A method as claimed in claim 7 wherein step d) of the method comprises identifying the presence of approximately 20 contiguous nucleotides of the nucleic acid molecule comprising SEQ ID NO. 2 or a complement thereof.

18. A method as claimed in claim 7 wherein step c) utilizes one oligonucleotide primer complementary to 12 contiguous nucleotides of SEQ ID NO. 2 or a complement thereof; and one oligonucleotide primer complementary to 10-12 nucleotides of IS*900* or a complement thereof.

19. A method as claimed in claim 7 wherein step c) utilizes one oligonucleotide primer complementary to substantially 15 contiguous nucleotides of SEQ ID NO. 2 or a complement thereof; and one oligonucleotide primer complementary to substantially 15 nucleotides of IS*900* or a complement thereof.

20. A method as claimed in claim 7 wherein step c) utilizes one oligonucleotide primer complementary to substantially 20 contiguous

nucleotides of SEQ ID NO. 2 or a complement thereof; and one oligonucleotide primer complementary to substantially 20 contiguous nucleotides of IS*900* or a complement thereof.

21. The use of a probe comprising substantially 10-12 contiguous nucleotides selected from the nucleic acid comprising SEQ ID NO. 2 or a complement thereof to determine whether a strain of either sheep type or cattle type *M. paratuberculosis* is present in a sample.

22. The use of a probe comprising at least 15 contiguous nucleotides selected from the nucleic acid comprising SEQ ID NO. 2 or a complement thereof to determine whether a strain of either sheep type or cattle type *M. paratuberculosis* is present in a sample.

23. The use of a probe comprising at least 20 contiguous nucleotides selected from the nucleic acid comprising SEQ ID NO. 2 or a complement thereof to determine whether a strain of either sheep type or cattle type *M. paratuberculosis* is present in a sample.

24. The use of SEQ ID NO.1 and/or SEQ ID NO. 2, or a fragment or complement thereof, to determine whether a strain of either a sheep type or a cattle type of *M. paratuberculosis* is present in a sample.

25. The use of SEQ ID NO.1, or a fragment or complement thereof, to distinguish any strain of *M. paratuberculosis* from any other strain of the MAI complex which may be present in a sample.

26. The use of SEQ ID NO.2, or a fragment or complement thereof, to distinguish any strain of *M. paratuberculosis* from any other strain of the MAI complex which may be present in a sample.

27. The use of SEQ ID NO.1, or a fragment or complement thereof, to distinguish any strain of *M. paratuberculosis* from any strain of the *M. tuberculosis* complex which may be present in a sample.

28. The use of SEQ ID NO.2, or a fragment or complement thereof, to distinguish any strain of *M. paratuberculosis* from any strain of the *M. tuberculosis* complex which may be present in a sample.

29. The use of SEQ ID NO. 1, or a fragment or complement thereof, to detect the presence of *M. paratuberculosis* as a causative agent of Johne's disease or Crohn's disease.

30. The use of SEQ ID NO. 2, or a fragment or complement thereof, to detect the presence of *M. paratuberculosis* as a causative agent of Johne's disease or Crohn's disease.